United States Department of Agriculture Animal and Plant Health Inspection Service Center for Veterinary Biologics P. O. Box 844 Ames, IA 50010

- 1. Reagent Name: Leptospira icterohaemorrhagiae Challenge Culture
- 2. Strain or Source: CF-1, NVSL No. 11403
- **3. Lot Number:** Identified by date of preparation.
- **4. Fill Date:** Cultures are prepared every 6 to 16 weeks from *L. icterohaemorrhagiae*-infected hamsters.
- **5. Expiration Date:** Use within 1 year from labeled date.

Precautions: *Leptospira* spp. are classified as a biosafety level 2 pathogen. Handle under a biological safety hood while wearing gloves. Use face shield when inoculating animals.

- **6. Intended Use:** Challenge material for *L. icterohaemorrhagiae* testing.
- 7. **Instructions for Use:** Upon receipt, subculture in fresh semisolid P80-BA medium before inoculating hamsters as described in Supplemental Assay Method (SAM) 610. *L. icterohaemorrhagiae* should be transferred through hamsters a minimum of three times before initiating a potency test according to **SAM 610**.
- **8. Test of Reagent:** Cultures are obtained from infected hamsters challenged with *L. icterohaemorrhagiae* according to **SAM 610**. Cultures are negative for extraneous bacteria when incubated on blood agar plates at 37°C. Cultures are checked for serological identity.
- **9. Container Size, Type, Weight, or Volume:** 16-mm x 125-mm screw-capped tubes containing 10 mL P80-BA semisolid medium with viable *L. icterohaemorrhagiae*.
- **10. Storage Conditions:** Store at room temperature (20°- 25°C). After subculture, store at room temperature after dinger zones (foci of leptospiral growth) are apparent.
- **11. CVB Technical Contact:** Bacteriology Section, Center for Veterinary Biologics, (515) 337-6140 or FAX (515) 337-7673.
- **12. Origin and Passage History:** Origin: Puerto Rico, dog isolate. Walter Reed Army Institute of Research through a commercial laboratory (1969). This strain is hamster virulent.

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13. Method of Preparation: Liver tissue and kidneys from an *L. icterohaemorrhagiae*-infected hamster are homogenized and diluted in phosphate-buffered saline containing 1% bovine serum albumin. Dilutions of the tissue suspension are inoculated onto semisolid P80-BA medium and incubated at 28°- 30°C for 2-4 weeks, or until dinger zones are apparent. Cultures are stored at room temperature (20°- 25°C) after dinger zones appear.

14. Other: None.

Reagent orders and feedback should be sent *including phone number* to the following email address: CVB@aphis.usda.gov

Reagent orders forms (APHIS 2018) are available from: http://www.aphis.usda.gov/animalhealth/cvb forms

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